Historic, archived document

Do not assume content reflects current scientific knowledge, policies, or practices.

SHOUT
SHARE
IT

UNITED STATES DEPARTMENT OF AGRICULTURE

BULLETIN No. 581

Contribution from the Bureau of Chemistry
CARL L. ALSBERG, Chief

Washington, D. C. ▼ October 6, 1917

MICROSCOPICAL STUDIES ON TOMATO PRODUCTS.

By BURTON J. HOWARD, *Microscopist in Charge, in collaboration with* CHARLES H. STEPHENSON, *Scientific Assistant, Microchemical Laboratory.*

CONTENTS.

	Page.
Introduction	1
Relation between visual inspection and microscopical examination	2
Tomato pulp	2
Catsup	5
Products made from peeled stock	7
Relation between percentage of rot and microscopical counts	8
Mold counts	8
Laboratory samples	8
Factory samples	11
Spore and yeast counts	14
Relation between percentage of rot and microscopical counts—Continued.	
Bacterial counts	16
Microscopic counts on tomato sauces and pastes	16
Method for microanalysis of tomato products	21
Apparatus	21
Molds (tentative)	21
Yeasts and spores (tentative)	22
Bacteria (tentative)	22
Explanation of calculations	23
Summary	24

INTRODUCTION.

During the past few years the Bureau of Chemistry has conducted a very comprehensive investigation to establish a basis for judging tomato products. Experiments were conducted in the bureau laboratory and also in factories, a large number of which were visited. Out of the mass of data thus collected, it is felt that the scientific facts underlying the relationship between microorganisms and the rot and decay of tomato products should be of value to manufacturers and food control officials at this time. The results bearing upon the relation of the physical condition of the stock from which tomato products are made to the number of microorganisms present in tomato products are therefore given in this bulletin.

A great many tests at factories were made by noting the general condition of the stock and then examining microscopically samples of the finished product. The criteria published in 1911 in Bureau of Chemistry Circular 68,[1] for the guidance of manufacturers, were

[1] Sold by U. S. Superintendent of Documents Washington, D. C., for 5 cents.

reached largely by such experiments. At that time very few tests had been made to correlate the amount of rot by weight with the microscopical counts. In the present discussion, therefore, the results obtained by visual inspection are considered first. The other series, that in which the percentage of rot was estimated, is discussed later (p. 8).

RELATION BETWEEN VISUAL INSPECTION AND MICROSCOPICAL EXAMINATION.

Tables 1 to 6, inclusive, and Tables 9 to 12, inclusive, show that a fairly concordant relationship exists between the character of stock as determined by visual inspection and the microscopic counts on the final product. In Bureau of Chemistry Circular 68 it was suggested that by good, sanitary practice the mold count could be kept down to 25 per cent or less, the yeast and spore count to 25 or less, per 1/60 cubic millimeter and the bacteria per cubic centimeter to 25 million or less. In only one instance in Tables 1 to 6 (Table 1, S 32) are these suggested limits exceeded on stock which to visual inspection appeared acceptable. In this sample the bacteria ran somewhat higher than is usual for passable stock. These results show that good stock properly handled almost never runs high in microorganisms.

In general, high counts of microorganisms are obtained on products made from stock which by visual inspection would be judged as objectionable or from stock which has been improperly handled. There seem to be, however, more exceptions to this rule than to that governing the microorganism count on products made from good stock. In three or four samples the counts are comparatively low on stock which to visual inspection appeared dubious. The few such exceptions which from time to time occur do not modify the fact that high counts of organisms indicate unmistakably that the stock used was in bad condition or that it was handled in an insanitary manner during manufacture. Either of these causes furnishes sufficient ground for condemnation of the product.

TOMATO PULP.

The average of the counts on pulp from stock which appeared acceptable from a visual inspection (Table 1) is as follows: Molds in 8 per cent of the fields; yeasts and spores, 8 per 1/60 cmm; bacteria, 5,900,000 per cc. Table 2 gives the results of counts on pulp made from unacceptable or questionable stock. Taking averages of the counts in this table, excluding samples of questionable character, the following results are obtained: Molds, 51 per cent of the fields; yeasts and spores, 51 per 1/60 cmm; bacteria, 77,900,000 per cc.

TABLE 1.—*Analysis of pulp made from stock approved by visual inspection.*

Sample.[1]	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
H 61a	Pulp, skin and core; fairly well sorted and made up promptly	O. K.	9	(3)	16
H 61b	.....do	O. K.	11	17	12
H 73b	Pulp, skin and core; well sorted and made up promptly	O. K.	13	9	20
H 77a	Pulp, whole tomatoes; sorted under personal supervision	O. K.	5	7	4.5
H 88	Pulp, whole tomatoes; well washed and sorted	O. K.	2	3	11
H 89	Pulp, skin and core; well washed and sorted	O. K.	9	10	12
H 90	Pulp, whole tomatoes; well washed and sorted	O. K.	5	4	5
H 91	Pulp, skin and core; well washed and sorted	O. K.	15	8	15
H 104	Pulp, whole tomatoes; tomatoes ran good and then were sorted	O. K.	2	5	5
H 105	.....do	O. K.	2	5	7
H 279	Pulp, whole tomatoes; stock poor (many decayed), but well washed and sorted	O. K.	6	3	6
H 283	.....do	O. K.	6	14	12
S 32	Whole-tomato pulp	O. K.	7	33	2
S 44	Whole-tomato pulp from cyclone	O. K.	18	16	2
S 45	.....do	O. K.	13	24	12
S 54	.....do	O. K.	8	17	22
S 57	Whole-tomato pulp from cyclone; tomatoes sorted	O. K.	11	8	8
S 60	.....do	O. K.	8	12	6
S 62	.....do	O. K.	12	8	6
S 67	.....do	O. K.	3	5	10
S 68	Whole-tomato pulp from finisher; tomatoes sorted	O. K.	7	(3)	8
S 70	Whole-tomato pulp; tomatoes sorted	O. K.	12	9	4
S 76	Whole-tomato puree; tomatoes sorted	O. K.	11	9	8
S 77	Whole-tomato pulp; tomatoes sorted	O. K.	4	9	4
S 79	.....do	O. K.	6	8	4
S 81	.....do	O. K.	15	10	6
S 85	Whole-tomato puree from finisher	O. K.	9	11	2
S 87	Whole-tomato pulp; tomatoes sorted and well washed	O. K.	4	4	2
S 88	.....do	O. K.	3	3	.....
S 93	Whole-tomato pulp; tomatoes sorted	O. K.	6	6	4
S 97	.....do	O. K.	6	5	8
S 99	.....do	O. K.	14	9	10
S 107	Whole-tomato pulp; indifferent sorting	O. K.	4	10	10
S 108	.....do	O. K.	.....	.....	14
S 110	Whole-tomato pulp; well sorted	O. K.	6	2	8

[1] H, analyzed by B. J. Howard; S, analyzed by C. H. Stephenson.
[2] O. K., good, or acceptable.
[3] Very few.

Table 2 also serves to show that delay during the manufacture of pulp usually produces a rapid increase in the growth of organisms, noticeably yeasts and spores, or bacteria, or both, with the result that high counts are obtained for these organisms in the final product. This is well illustrated in Samples H 59, H 67, H 96a, and S 64. The absolute necessity of handling tomato products promptly in order to avoid secondary spoilage is shown in the case of Samples S 70 and S 78. Sample S 70 is the count on a sample of pulp freshly made, while Sample S 78 represents the same batch after standing 24 hours. The counts are:

	S 70	S 78
Fields with molds (per cent)	4	4
Yeasts and spores (per 1/60 cmm)	12	9
Bacteria (million per cc.)	9	218

TABLE 2.—*Analysis of pulp made from bad or questionable stock as determined by visual inspection.*

Sample.[1]	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
H 57	Puree, skin and core; very little sorting	C	45	[3]	18
H 58	Pulp, skin and core; poorly sorted	C	15	[3]	75
H 59	Pulp, skin and core; apparently well sorted but stock left in barrels Saturday to Tuesday	C	162	240	10
H 63	Puree, skin and core; fairly well sorted and made up promptly	?	32	12	20
H 67	Pulp, skin and core; very imperfectly sorted, allowed to stand 3 or 4 hours before pulping	C	60	192	67
H 70	Pulp, skin and core; very imperfectly sorted, allowed to stand 6 hours before pulping	C	35	44	36
H 73e	Pulp, skin and core; decayed condition, largely dry rot (laboratory experiment)	C	61	48	100
H 75b	Pulp, skin and core; indifferently sorted; allowed to stand 2 to 6 hours before pulping	C	55	50	22
H 76a	Pulp, whole tomatoes; fairly well sorted	?	7	5	5
H 80a	Pulp, whole tomatoes; fairly well sorted but open to criticism; some decayed tomatoes	?	7	9	13
H 81a	....do	C	18	12	17
H 83a	Pulp, whole tomatoes; sorting open to strong criticism	C	13	5	17
[4] H 84c	Pulp, skin and core; sorting wholly inadequate	C	63	60	100
[4] H 85	....do	C	114	120	92
[4] H 86	....do	C	150	84	100
[4] H 87	....do	C	155	84	100
H 92	Pulp, whole tomatoes; no sorting; washing not satisfactory	C	21	5	44
H 93	....do	C	15	7	45
H 94c	Pulp, whole tomatoes (gravity method); not sorted; too many damaged tomatoes	C	18	7	32
H 95a	Pulp, skin and core; sorting not wholly satisfactory (not promptly made up)	C	20	12	30
H 96a	Pulp, skin and core; prepared as 95a but left over night before pulping	C	61	200	29
H 97c	Pulp, skin and core; no sorting; bad conditions throughout; fairly prompt handling	C	37	26	65
H 98b	....do	C	72	43	90
H 103a	Pulp, whole tomatoes; sorting unsatisfactory	C	8	7	25
H 106a	Pulp, whole tomatoes; not sorted, acknowledged by manufacturer to need sorting	C	12	5	21
[5] H 108a	Pulp, skin and core; not sorted; all refuse from tables used; bad	C	17	7	26
[5] H 109	Pulp, skin and core; not sorted; all refuse from tables from day's accumulation kept hot until used	C	27	19	19
H 110b	Pulp, skin and core; all refuse with decayed parts from table used; bad	C	13	36	40
H 113a	Pulp, skin and core; all refuse from tables used; product condemned by employees	C	36	20	85
H 114a	....do	C	19	41	71
H 273	Pulp, skin and core; all refuse from tables used; no sorting; bad	C	31	41	74
H 274	....do	C	35	36	65
H 275	....do	C	42	53	86
H 276	....do	C	53	52	87
H 277	....do	C	44	52	84
S 22	Pulp, skin and core; all refuse from table used; objectionable	C	21	9	26
S 23	Skin and core pulp; tomatoes not sorted	C	25	13	40
S 24	....do	C	66	24	42
S 49	Tomato pulp from unsorted tomato peelings	C	61	66	37
S 64	Whole-tomato pulp, after standing 20 hours	C	42	773	6
S 78	Whole-tomato pulp, after standing 24 hours	C	9	218	4
S 80	Tomato waste from sorting tables made into pulp	C	270	365	80
S 102	Skin and core pulp; no sorting of tomatoes	C	21	28	45
S 103	Whole-tomato pulp; small tomatoes	?	16	21	26
S 109	Whole-tomato pulp; indifferent sorting	?	.....	.....	22

[1] H, analyzed by B. J. Howard; S, analyzed by C. H. Stephenson.
[2] C, bad product from inspection of raw material; ?, doubtful or open to criticism.
[3] Very few.
[4] Taken at same plant at different times the same day.
[5] Very little washing of tomatoes at start; were fairly good, but some decayed.

BARREL PULP.

Table 3 gives the counts obtained upon a number of samples of barrel pulp. The character of the stock from which these pulps were made is not known, beyond the fact that nearly all were skin and core pulps. The results, however, are of interest in comparison with the data in Tables 1 and 2. Table 3 brings out strongly the fact that in barrel stock the counts on yeasts and spores, and especially on bacteria, are usually very high. It also serves to explain the counts obtained on catsup made from barrel stock. In two instances the visual inspection of the barrel stock left a doubt as to whether the product was suitable for use, but such cases are rare. Excluding these two from the calculation, the averages of the counts on the remainder of the samples in this series are: Molds in 48 per cent of the fields; yeasts and spores, 77 per 1/60 cmm; bacteria, 207,700,000 per cc. The results in this table prove the inadvisability of trying to use barrels for storing tomato pulp.

TABLE 3.—*Analysis of pulp stored in barrels (condition of stock when made, not known.)*

Sample.[1]	Description.	Character from inspection when opened.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
H 16	Pulp in barrels, 7–9 months; odor of spoilage	C	22	96	66
H 17	Pulp in barrels, 7–9 months; odor of spoilage; gas	C	93	84	100
H 20	Pulp in barrels, 7–9 months; odor bad	C	160	408	50
H 22	Pulp in barrels, 7–9 months; odor bad; gas	C	52	168	98
H 27	do	C	52	320	98
H 28	do	C	122	340	33
H 29	do	C	250	312	[3]
H 32	Pulp in barrels, 7–9 months; very strong odor; bad; gas	C	45	144	40
H 33	Pulp in barrels, 7–9 months; odor bad	C	42	264	50
H 34	Pulp in barrels, 7–9 months; odor mild for barrel pulp	[5]	12	32	41
H 35	Pulp in barrels, 2 to 3 years; old; strong odor; skin and core	C	70	216	50
H 36	Pulp in barrels, 7–9 months; strong odor	C	130	228	40
H 37	Pulp in barrels, 7–9 months; strong odor; skin and core	C	32	192	50
H 38	do	C	21	180	33
H 39	Pulp in barrels, 7–9 months; odor of spoilage milder than usual; whole tomatoes	C	10	120	14
H 40	do	C	67	132	[4]
H 41	do	C	56	120	50
H 46	Pulp in barrels, 7–9 months; odor of spoilage milder than usual; one of the best seen	( )	15	29	[6]

[1] H, analyzed by B. J. Howard.
[2] C, bad product from inspection when opened.
[3] Few.
[4] Very few.
[5] Doubtful or open to criticism.
[6] Not determined.

CATSUP.

The results of counts of catsups (Tables 4 and 5) show that a relationship similar to that found in the case of pulps exists between products made from acceptable stock and those from decayed material. The counts on the good stock catsups averaged: Molds in 9 per cent of the fields; yeasts and spores, 11 per 1/60 cmm; bacteria, 8,900,000 per cc. Table 5 includes some catsups made from stock of doubtful character in addition to those that were unquestionably bad. If the doubtful ones are excluded from the calculation and an average taken of the remainder, the following approximate figures

are obtained: Mold in 50 per cent of the fields; yeasts and spores, 56 per 1/60 cmm; bacteria, 131,100,000 per cc. Attention should be called to the fact that the bacterial count was especially high because Sample H 284 was actively spoiling at the time of examination.

TABLE 4.—*Analysis of catsup made from stock approved by visual inspection.*

Sample.[1]	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
H 77b	Catsup, whole tomatoes; sorted under personal supervision	O. K.	12	12	12
H102b	Catsup, whole tomatoes; well sorted; made up promptly	O. K.	5	2.5	13
H107b	Catsup, whole tomatoes; well sorted	O. K.	10	7	9
H 272	Catsup, whole tomatoes; apparently good method used	O. K.	15	14	12
H 278	Catsup, whole tomatoes; poor stock, but well washed and sorted	O. K.	10	12	14
H 280	.....do	O. K.	8	7	11
H 281	.....do	O. K.	7	5	8
H 285	Catsup, whole tomatoes; tomatoes poor, many decayed when received; but well washed and sorted	O. K.	19	16	14
H 287	.....do	O. K.	10	12	11
H 288	.....do	O. K.	16	21	10
S 30	Catsup taken from filler; made from sorted tomatoes	O. K.	12	16	12
S 31	Catsup, whole tomatoes; tomatoes poor; many decayed when received; but well washed and sorted	O. K.	11	10	6
S 46	Whole tomato catsup	O. K.	10	14	14
S 58	Finished whole tomato catsup	O. K.	12	5	10
S 63	Catsup sauce for beans	O. K.	11	9	6
S 69	Finished whole tomato catsup; tomatoes sorted	O. K.	8	[3]	0
S 73	.....do	O. K.	6	3	10
S 74	.....do	O. K.	14	8	6
S 75	.....do	O. K.	7	4	4
S 83	.....do	O. K.	9	8	10
S 90	Whole tomato catsup; tomatoes sorted	O. K.	3	2	0
S 94	.....do	O. K.	20	6	6
S 98	.....do	O. K.	10	7	20
S 104	.....do	O. K.	24	10	16
S 105	.....do	O. K.	14	10	4
S 106	.....do	O. K.	11	13	8

[1] H, analyzed by B. J. Howard; S, analyzed by C. H. Stephenson.
[2] O. K., acceptable from inspection of raw material.
[3] Very few.

TABLE 5.—*Analysis of catsup made from bad or questionable stock as determined by visual inspection.*

Sample.[1]	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
H 71	Catsup, skin and core; poorly sorted; not made up promptly; stock stood 1–6 hours	C	89	240	21
H 72c	Catsup, skin and core; pieces more or less decayed; not promptly made	C	143	130	90
H 76c	Catsup, whole tomatoes; fairly well sorted, but not fully approved	[3]	22	15	20
H 80b	Catsup, whole tomatoes; fairly well sorted, but not approved	[3]	15	26	25
H 81b	Catsup, whole tomatoes; sorting open to criticism; not effective	[3]	20	24	34
H 82b	Catsup, whole tomatoes; sorting carelessly done and ineffective	C	34	17	52
H 83b	.....do	C	40	17	61
H 103b	Catsup, whole tomatoes; imperfectly sorted; made up promptly	[3]	13	12	37
H 106b	Catsup, whole tomatoes; not sorted but needing it; made promptly, well washed	C	19	7	36
H 111c	Catsup, skin and core; conditions very bad, refuse of every kind used without sorting; gravity method	C	55	91	81
H 282	Catsup, whole tomatoes; fairly well sorted and washed	[3]	15	35	16
H 284	Catsup, from barrel; vigorously fermenting	C	10	416	9

[1] H, analyzed by B. J. Howard.
[2] C, condemned in opinion of writers.
[3] In doubt and at least open to criticism.

PRODUCTS MADE FROM PEELED STOCK.

A few counts on chili sauce are given in Table 6. The tomatoes used for this were a poor grade and required extensive trimming to remove the decayed material. The counts indicate how well this work was being done and what may be regarded as the upper limit for products made from peeled stock such as is used in chili sauce manufacture.

TABLE 6.—*Analysis of chili sauce.*[1]

Sample.	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 160 cmm.	*Million per cc.*	*Per cent.*
72d	Poor raw material, well trimmed. (Catsup No. 72c made from trimming of these)	O. K.	11	12	2.5
73a	Raw stock well sorted. (Trimmings made into pulp 73b)	O. K.	4	2	[3]
73aa	Raw stock partly decayed, well trimmed	O. K.	6	9	[4]
75a	From sorted tomatoes	O. K.	1	5	2
84d	Stock. (Peeled tomatoes from which trimmings for pulp 84c was obtained)	O. K.	6	5	9

[1] Analyses by B. J. Howard.
[2] O. K., acceptable from inspection of raw material.
[3] None in 58 fields.
[4] None in 39 fields.

These figures are rather high for canned peeled tomatoes. Well handled stock usually gives very low counts. Fermented stock or reprocessed swells as a rule show a marked increase in organisms other than molds. Molds occasionally develop, however, to a noticeable degree in sealed cans, although such instances are rare. One remarkable example of this character came to the notice of the bureau several years ago. A shipment of pulp in 5-gallon cans was condemned because masses of mold, some as large as a man's head, were found in a number of cans. The size of many of the masses precluded any possibility of their having been present at the time the cans were filled.

The bureau is sometimes asked whether it is advisable to sort raw stock intended for peeling, provided the trimmings are to be discarded. In answer to this question attention might be called to two or three of the chili sauce samples given in Table 6 and the products made from their trimmings. As commonly understood, chili sauce is made from peeled tomatoes handled as for canning. The skins, cores, and bad portions are supposed to be removed during peeling. Chili sauce, Sample 72d, was made from poor, rather badly decayed stock, which, however, was so well trimmed that it was acceptable. The same was true of the stock from which Sample 84d was made. The stock used for chili sauce, Sample 73a, while poor, was well sorted before being peeled. Although a comparison of these three samples shows the counts to be slightly lower in the case of the sorted stock, on the whole the advantage gained scarcely justifies making sorting a requirement on general stock if the peeled tomatoes are the only portions to be used. In these particular cases, the trimmings

were used for other products. The following comparison of the counts on them is, therefore, of interest:

	Fields with molds.	Yeasts and spores.	Bacteria.
	Per cent.	*Per 1/60 c.mm.*	*Million Per cc.*
Catsup made from trimmings of tomatoes peeled for 72d	90	143	130
Pulp from trimmings of tomatoes peeled for 84d	100	63	60
Pulp from trimmings of well-sorted tomatoes peeled for 73a	20	13	9

The contrast between the character of the first two and the last is a good illustration of the counts obtained on such types of products handled as these were.

RELATION BETWEEN PERCENTAGE OF ROT AND MICROSCOPICAL COUNTS.

During the earlier investigations made upon tomato products the character of the stock was usually determined by means of visual inspection only. In later experiments the counts were made on products in which the amount of rot in the raw stock was determined by weight. These experiments were divided into two groups. One group of samples was prepared in the laboratory from stock of known character, while the other group consisted of inspected stock made in the factory. "Decayed" or "rotten" stock is taken to mean such portions as would appeal to the average intelligent housewife or consumer as being decayed or rotten and unfit or undesirable for food. For purposes of study of the molds the two groups are considered separately.

MOLD COUNTS.

LABORATORY SAMPLES.

In tests conducted in the laboratory the amount of rot in the whole tomato was determined by cutting out and weighing the decayed portion and calculating the percentage. These portions were then mixed and pulped by rubbing through a 20-mesh sieve. In a few cases this procedure was modified by pulping the good and bad portions separately and then mixing them in the proportions desired. Experience has shown, however, that such a pulp is usually not quite as satisfactory to work with as the products normally manufactured because it is somewhat difficult to produce in the laboratory a pulp of just the same texture as that made under good factory conditions. The experiments and counts are nevertheless incorporated in this bulletin, because when compared with the counts on pulp made under factory conditions they show that the same general relationship between the percentage of rot and counts is maintained. The

results obtained (Table 7) are tabulated in the order of increasing amounts of decay or rot present in the original stock. Samples containing 1 per cent or less of rot show low counts on each type of organism, while in those containing more than that amount one or another of the organisms usually runs higher than is the case in good stock.

TABLE 7.—*Relation between percentage of rot and microscopical count on laboratory samples.*

Sample.	Rot, by weight, in stock.	Fields with molds.	Yeasts and spores.	Bacteria.	Sample.	Rot, by weight, in stock.	Fields with molds.	Yeasts and spores.	Bacteria.
	Per cent.	*Per cent.*	*Per 1/60 cmm.*	*Million per cc.*		*Per cent.*	*Per cent.*	*Per 1/60 cmm.*	*Million per cc.*
1	0	0	4	5	24	9.5	16	25	18
2	0	2	4	4	25	9.9	36	14	83
3	0	0	9	3	26	10.0	42	19	123
4	0	0	3	2	27	10.0	42	19	116
5	0	2	4	3	28	10.4	30	41	93
6	0	0	3	2	29	11.7	6	68	87
7	0	0	2	5	30	12.4	54	23	34
8	0	0	3	2	31	13.8	32	35	32
9	0	0	9	14	32	15.0	57	22	197
10	1	2	20	10	33	18.0	20	31	29
11	1.8	0	20	28	34	19.1	66	9	223
12	2	8	27	18	35	20.0	70	24	402
13	3	8	23	29	36	20.0	63	22	366
14	3.4	8	1	2	37	22	38	31	42
15	4	16	35	55	38	23.6	50	34	101
16	5	22	38	69	39	24.3	74	12	9
17	5	4	28	13	40	31	42	37	49
18	5	22	20	65	41	40	64	34	54
19	5.0	27	16	76	42	50.9	64	36	60
20	5.2	12	12	28	43	100.0	98	210	396
21	5.5	50	7	5	44	100.0	100	35	65
22	7.3	0	6	93	45	100.0	100	22	960
23	7.5	24	4	15	46	100.0	98	103	860

No high mold count occurs in samples low in amount of rot (Table 7). In some instances, however, low counts of mold were obtained in samples containing a substantial amount of rot. In such cases the decay was due principally to yeasts or bacteria, the count for one of which is usually high (Samples 18, 22, and 29). If, however, mold is the principal cause of decay, the counts on yeasts and bacteria are low (Samples 21 and 39).

In Figure 1 the mold counts have been plotted as ordinates and the percentage of rot in the stock as abscissas. Having plotted on the chart the mold counts in Table 7, it is possible by connecting the outlying points to define the approximate limits of what may be called the "Zone of Possible Mold Counts." From the chart it is seen that within this zone any count within certain limits may occur. In the writer's experience no counts on laboratory samples beyond these limits have been observed. For instance, no laboratory sample with less than 5.5 per cent of rot gave a mold count of more than 50. In making the chart the zone beyond 24.3 per cent of rot is represented on the basis of Samples 44 and 45, though the chart as drawn does not extend beyond the point of 30 per cent rot.

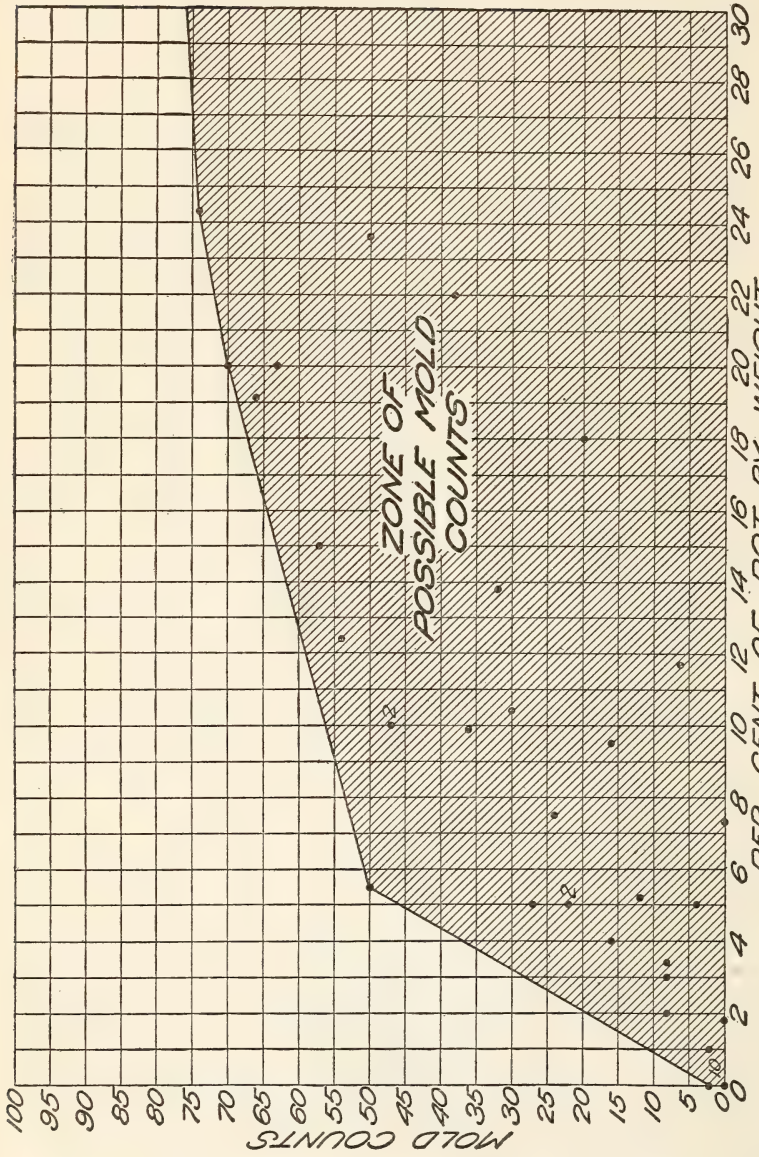

FIG. 1.—Relation between percentage by weight of rot and mold count (laboratory samples).

FACTORY SAMPLES.

To obtain data on a practical manufacturing scale, tests under various factory conditions were conducted. In some of the plants the conditions were excellent, while in others they were highly objectionable. In making the tests the amount of decay was estimated or determined by weight as accurately as was practicable. With whole tomato stock it was usually customary to take a representative sample of from 25 to 50 pounds from the stream of sorted and washed stock on its way to the breaking-up tanks or to the cyclones. Each tomato in such a sample was inspected critically and the bad portions cut out and separated from the good portions. The parts thus separated were then weighed and the percentage of rotten or decayed material calculated. To estimate the amount of decay in the trimming pulps the amount of decayed material in the peeling stock was determined as in the case of the whole tomato stock. Since all the decayed material in the peeling stock is left in the trimmings, the percentage as determined on the scalded stock was multiplied by two. Although this may not be strictly accurate, it approximates the true figure nearly enough to be used as a practical working factor. It was adopted in calculating the data after tests had been conducted in various plants to determine what proportion of the stock sent to the peelers is left in the trimmings.

Field work performed during the last three seasons has demonstrated that with proper equipment and factory management there is no excuse for the stock which is ready for the cyclone to contain over 1 per cent of decayed material. Any excess is traceable to some fault in the system or to improper management.

TABLE 8.—*Relation between amount of rot and microscopical counts on tomato products, factory samples.*[1]

Day.	Average rot, by weight, in stock.	Average fields with molds.	Average yeasts and spores.	Average bacteria.	Day.	Average rot, by weight, in stock.	Average fields with molds.	Average yeasts and spores.	Average bacteria.
	Per cent.	*Per cent.*	*Per 1/60 cmm.*	*Million per cc.*		*Per cent.*	*Per cent.*	*Per 1/60 cmm.*	*Million per cc.*
1	0.2	5	9	9	21	1.0	20	.....	.....
2	0.2	6	.....	.....	22	1.2	13	4	6
3	0.3	7	.....	.....	23	1.4	13	.....	.....
4	0.4	13	.....	.....	24	1.8	9	7	9
5	0.4	7	6	9	25	1.9	29	.....	.....
6	0.5	5	.....	.....	26	2.0	13	.....	.....
7	0.5	9	.....	.....	27	2.1	24	.....	.....
8	0.5	21	.....	.....	28	2.2	16	.....	.....
9	0.5	11	3	5	29	2.4	16	.....	.....
10	0.6	21	.....	.....	30	3.2	35	.....	.....
11	0.8	16	4	9	31	3.2	50	.....	.....
12	0.8	24	8	15	32	3.8	38	13	14
13	0.8	18	14	10	33	3.8	17	.....	.....
14	0.9	19	11	9	34	4.0	41	16	14
15	0.9	20	13	7	35	4.0	60	.....	.....
16	[2]	9	.....	.....	36	6.0	32	11	11
17	[2]	14	.....	.....	37	7.8	49	.....	.....
18	1.0	2	2	8	38	9.8	72	.....	.....
19	1.0	8	5	6	39	20.0	87	64	156
20	1.0	12	.....	.....					

[1] Summary of all determinations on similar products made on same day. [2] Under 1.0.

Table 8 summarizes 39 days' work under factory conditions on the relation between the amount of rot and the microscopical counts. As in Table 7, the results are arranged in the order of the amount of decayed material in the original stock. Under factory conditions it is frequently difficult to trace a definite portion of raw stock through the whole process of manufacture, with any certainty that the final product is from the same stock, unmixed with other lots. It has therefore been considered advisable to average the daily results obtained on the amount of rot and compare them with corresponding daily averages on the microscopic tests. It might be stated in this connection that the results thus summarized in Table 8 represent experiments conducted in 17 different factories and involve 179 determinations of percentage by weight of rot in stock and 235 mold counts.

Experience has shown that in factories where good, thorough washing is employed and where promptness of handling is observed the mold count is of greater importance in judging the condition of the raw stock than the counts on the other organisms. High yeast and spore, and bacterial counts usually indicate secondary spoilage. In order to secure as large a mass of data as possible on the mold content, complete counts for the other organisms were made in only a few cases during the field work of 1916. Hence the blanks in the table. A comparison of the field results shows that, as in the case of the laboratory tests, high mold counts were not found in the products made from stock which had been well handled and sorted.

The results given in Table 8 emphasize two points also brought out in Table 7: First, none of the samples having 1 per cent or less of rot give excessively high microscopical counts; second, with increasing amounts of rot more or less increase in one or another of the microorganisms occurs. They also show that while a comparatively low count may not always indicate a product made from suitable stock, it is clearly demonstrated that a high count on any one or more of the three types of organisms indicates bad stock. It appears, then, that one weak spot in the micro-counting system is its failure to disclose some bad products which should be condemned.

To facilitate the study, the mold counts given in Table 8 have been plotted graphically in Figure 2, in which the "Zone of Possible Mold Counts" is indicated by the shaded portion. Attention should be called to the fact that beyond 20 per cent of rot the chart is plotted on the basis of the assumption that 100 per cent rot would give a mold count of 100 per cent of the fields. As a matter of fact, the mold count reaches this maximum of 100 per cent of the fields with less than this amount of rot, as will be seen by reference to the laboratory series. These high counts, while occurring occasionally in commercial samples, are now rare. In the studies of the relationship

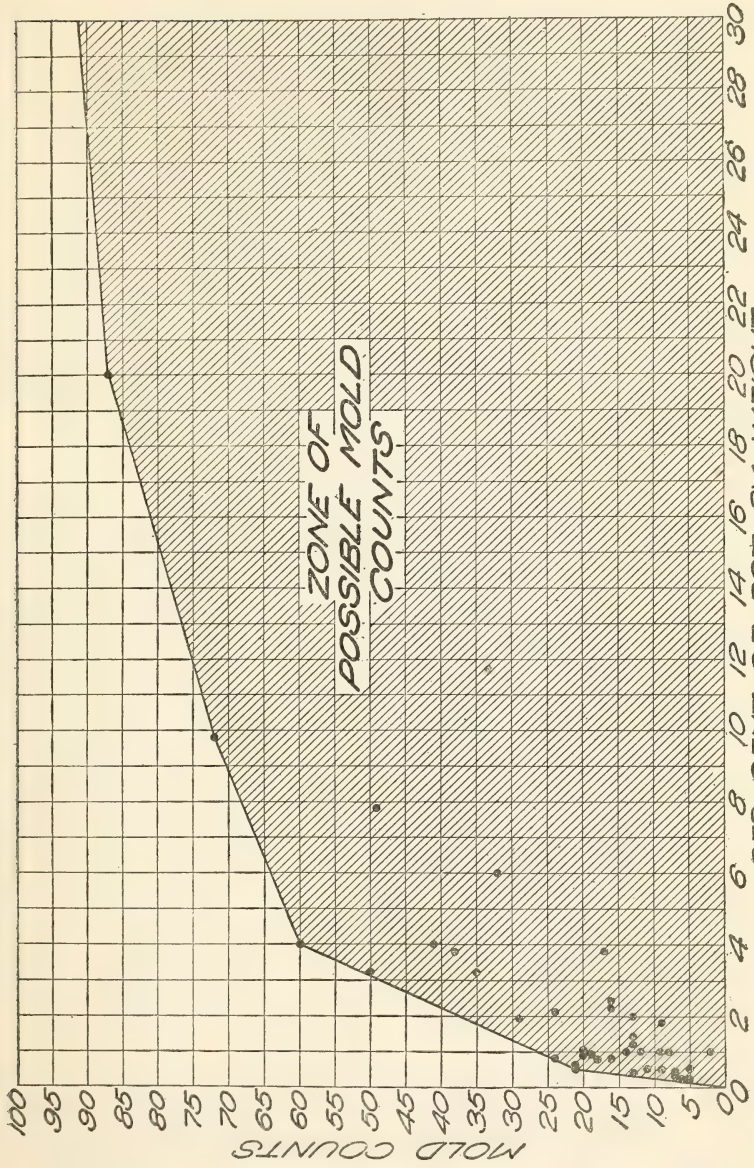

FIG. 2.—Relation between percentage by weight of rot and mold count (factory samples).

between rot and count under factory conditions, 20 per cent of rot was the highest on which the full data were secured. From 0 to ½ per cent of rot the mold count rises sharply. Beyond ½ per cent the rate of rise gradually decreases, until after 20 per cent of rot the rate of increase is slow. On the whole, the zone was higher on factory than on laboratory samples. The chart shows that a count of 60 per cent molds represents a rot content of not less than about 4 per cent. It is interesting to note that the mold count of 25, which was suggested as a factory working basis in 1911, represents at least 0.8 to 0.9 per cent of rot. There may be more rot than that present, but, on the basis of the data at hand, it is highly improbable that it represents less than that amount. From the chart it is possible to calculate the approximate minimum percentage of decay represented by a given mold count. Thus, for instance, a mold count of 40 enters the "Zone of Possible Mold Counts" at a point representing 2.2 per cent of decay. Therefore a count of 40 may be obtained in samples having any amount of rot between 2.2 and 100 per cent.

SPORE AND YEAST COUNTS.

As has been pointed out, such a degree of efficiency in washing and sorting may be maintained in good factory practice that the percentage of decay in stock will be 1 per cent or less. With this in mind, an examination of Tables 7 and 8 shows that on samples containing such amounts of objectionable material the counts on yeasts and spores are 20 or less per 1/60 cmm. In the majority of the samples the comparatively low numbers of this type of organisms are noteworthy. This fact serves to emphasize what has already been stated, that high counts of these organisms are more frequently an indication of secondary than of primary spoilage. Thus in the factory series of samples (Table 8), which includes those having large amounts of decay, only one sample high in these organisms is found. In this case (Sample 39) it was trimming stock and was not made up as promptly as it should have been; consequently a certain amount of secondary spoilage had occurred. The estimate of 20 per cent rot is therefore under rather than over the true amount. All of the other factory samples were handled in such a way as to prevent appreciable secondary spoilage.

The combined results of yeast and spore counts, both on laboratory (Table 7) and factory tests (Table 8), have been plotted in Figure 3. Here it is seen that up to a count of 10 the results may be regarded as negligible. A count of 20 represents about 1 per cent of decay. From this point the rate of increase is slower. As in the case of molds, the rule that a moderately low yeast and spore count may not always indicate good stock but does indicate at least a certain amount of rot, holds good for this class of organisms. It is also

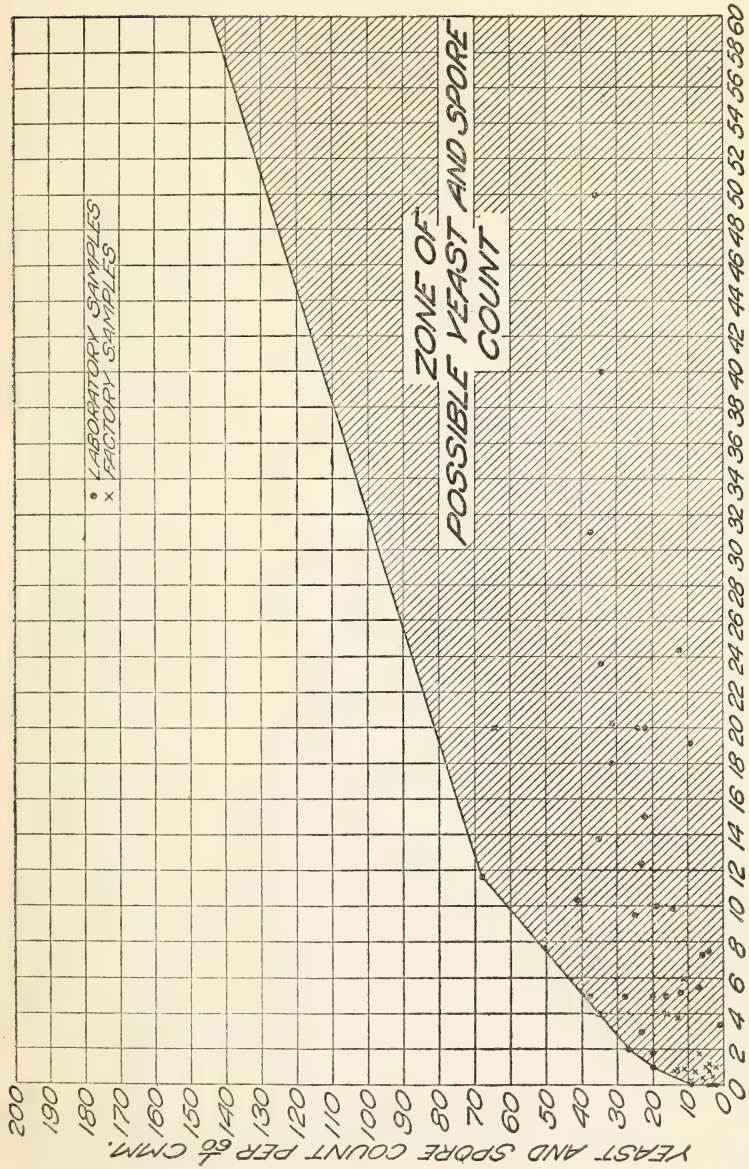
YEAST AND SPORE COUNT PER 66 CMM.
PER CENT OF ROT
ZONE OF POSSIBLE YEAST AND SPORE COUNT
• LABORATORY SAMPLES
× FACTORY SAMPLES

FIG. 3.—Percentage by weight of rot, and yeast and spore count. (The line showing the upper limit of "Zone of Possible Yeast and Spore Count" beyond 12 per cent of rot is based on sample 46, Table 7).

shown that high counts invariably indicate bad stock. From the chart it appears then that a count of 35 yeasts and spores in a unit volume may indicate 4 per cent or more of decayed material. By connecting the outlying points in the chart, the upper limits of the "Zone of Possible Yeast and Spore Counts" are obtained. The limit of the zone shown beyond 12 per cent of rot has been based on the count for laboratory Sample 46. From such a chart it is possible to calculate the minimum percentage of rot which a certain count represents.

BACTERIAL COUNTS.

Considering now the counts of bacteria in Tables 7 and 8, it is found that in the samples with 1 per cent or less of rot the count on bacteria is relatively low. With greater amounts of rot more bacteria may be found, till in Sample 45 a count of nearly a billion was obtained. In this case there was also a maximum mold count (100 per cent). Comparing the results in Table 7 with those in Table 8 it is noted that the mold count runs up more rapidly in the factory than in the laboratory series. With yeasts and spores and with bacteria, however, the reverse is true, that is, the counts mount more rapidly in the laboratory series. This is explained by the fact that in the laboratory series the tomatoes had not been subjected to the mechanical or factory method of washing, which results in eliminating much of the soft rot wherein these organisms are most abundant. The decay which resists this washing process is more commonly due to molds. The other organisms when present are usually, though not always, the result of secondary infection, and produce some of the very soft types of decay.

The results of bacterial counts on the laboratory and factory samples, tabulated in Tables 7 and 8, have been plotted in Figure 4. Below 15,000,000 bacteria the results indicate little as to the amount of decay, but above this point, up to about 20 per cent of rot, the ratio of increase as shown on the chart is about 20,000,000 for each per cent of rot. Beyond 20 per cent of rot the upper limit of the zone is based on the count on Sample 45 (Table 7). As in each of the other charts, the rule holds true here that a low bacterial count does not necessarily indicate sound stock, but a high bacterial count always indicates bad stock.

MICROSCOPIC COUNTS ON TOMATO SAUCES AND PASTES.

Tomato sauce and paste are practically the same kind of product as tomato pulp, but concentrated to a much greater degree. Hitherto no opinion has been published by the Bureau of Chemistry concerning the microscopic counts obtained on the more highly concentrated tomato products, when handled in accordance with good factory practice. During the season of 1912 a study of these products was

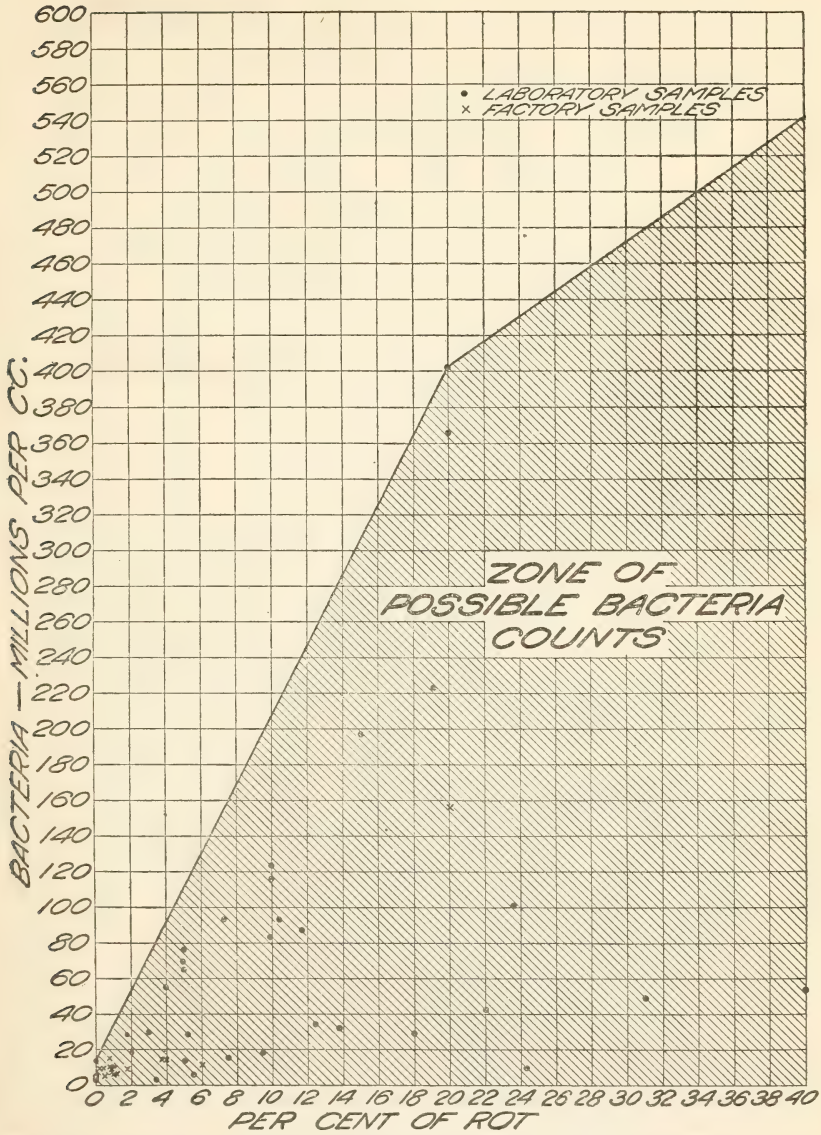

FIG. 4.—Percentage by weight of rot and bacterial count. (The line showing the upper limit of "Zone of Possible Bacteria Counts" beyond 20 per cent of rot is based upon the count on sample 45, Table 7),

made under factory conditions in Italy. These studies included investigation into the conditions of manufacture in each of the important tomato-producing sections of that country. In many of the factories experimental work was conducted. Tables 9 to 12 have been compiled from the data thus collected.

TABLE 9.—*Analysis of Italian tomato pulp made from stock approved by visual inspection.*[1]

Sample.	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
519	From freshly delivered tomatoes	O. K.	7	14	5
521	....do	O. K.	5	3	5
524	From fresh tomatoes	O. K.	5	[3]	8
528a	From freshly delivered tomatoes	O. K.	5	10	11
529	....do	O. K.	5	7	18
530	....do	O. K.	3	14	8
539a	....do	O. K.	8	7	9
548a	Made under personal supervision from freshly delivered tomatoes: washed but not sorted	O. K.	6	5	7
552a	From freshly delivered tomatoes: controlled experiment; washed but not sorted	O. K.	16	10	4
555a	From freshly delivered tomatoes: washed but not sorted	O. K.	11	5	2
596	From approved trimmings: controlled factory test	O. K.	3	5	1
602	Made under personal supervision from acceptable trimmings	O. K.	8	4	11

[1] Analyses by B. J. Howard. [2] O. K., acceptable from inspection of raw material. [3] Very few.

TABLE 10.—*Analysis of Italian tomato pulp made from bad or questionable stock as determined by visual inspection.*[1]

Sample.	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
523	From trimmings not made up promptly; fermenting	C	56	110	4
540a	From tomatoes stored in bin 48 hours; very sour and moldy; no washing or sorting	C	253	96	69
542	From tomatoes stored in bin about 54 hours; very sour and moldy; no washing or sorting	C	305	132	100
543a	....do	C	275	115	20
545a	From tomatoes stored in bin 3 or 4 days with 50 per cent fresh tomatoes	C	200	250	30
546	From tomatoes stored in bin 24 hours; in bad condition	C	164	53	44
549	From tomatoes washed and stored about 8 hours in bin; not acceptable	C	24	22	20
551a	From tomatoes stored in baskets; sour and moldy	C	136	25	31
553	From tomatoes stored in baskets 24 to 48 hours; bad condition	C	177	60	36
558	From tomatoes bin-stored, sour and moldy	C	109	43	19
573	From trimmings, waste and bad tomatoes	C	163	72	47
575	From trimmings obtained from peeling tables; not acceptable	C	131	58	63
577	From decayed tomatoes	C	175	72	76
578	....do	C	350	240	80
579	From trimmings taken promptly from tables but not of acceptable character	C	20	10	25
580	From trimmings allowed to stand 4 or 5 hours before pulping	C	85	60	24
592	From trimmings from fresh tomatoes, but delayed in pulping	C	31	45	19
595	From whole tomatoes; many decayed	C	53	50	60
617	From partly decayed stock	C	110	30	42
622	....do	C	170	25	44

[1] Analyses by B. J. Howard. [2] C, condemned; bad product from inspection of raw material.

Table 9, which gives the data on the unconcentrated juice, shows that on acceptable stock, as determined from visual inspection, the counts were low, ranging no higher than on American stock. The average counts on these samples are: Molds, 8 per cent of the microscopic fields; yeasts and spores, 8 per 1/60 cmm.; bacteria, 7,500,000 per cc. On the other hand, Table 10 shows the counts on unconcentrated pulps from bad or questionable stock. In this case there is a marked increase in the number of organisms, especially of yeasts and spores and bacteria over that found to occur in sound stock. Hence it is reasonable to conclude that with proper control over the manufacturing processes Italian products should be fully equal to American products made under similar conditions. The finished product is on an average 2 or $2\frac{1}{2}$ times more concentrated

TABLE 11.—*Analysis of Italian tomato sauces made from acceptable or fairly acceptable stock.*[1]

Sample.	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
520	From freshly delivered tomatoes; made up promptly	O. K.	24	12	15
528b	.....do.....	O. K.	25	38	18
531	.....do.....	O. K.	40	40	12
532	.....do.....	O. K.	46	28	15
536	.....do.....	O. K.	48	24	12
538	.....do.....	O. K.	32	40	11
539b	From freshly delivered tomatoes.....	O. K.	46	23	14
548b	From freshly delivered tomatoes; washed but not sorted; fairly good stock.....	O. K.	51	31	22
561	Made under personal supervision from fresh tomatoes.....	O. K	40	25	4

[1] Analysis by B. J. Howard. [2] O. K., acceptable from inspection of raw material.

than standard pulp. Table 11 shows that the molds do not exceed 25 per cent of the fields, or the counts obtained on normal pulp. On good stock the upper limit for yeasts and spores appears to be about 50 per 1/60 cmm., while for bacteria it is about 40 million per cc. In other words, the mold count does not seem to be much higher than that on the pulp, while the increase in the count for yeasts, spores, and bacteria is more nearly proportional to the degree of concentration. Examination of counts on the sauces and pastes made from objectionable stock as determined by visual inspection (Table 12) shows that as a class they run particularly high in yeasts and spores and bacteria and are also rather high in molds. The average of these counts, excluding the questionable samples, is: Molds, 57 per cent of the fields; yeasts and spores, 511 per 1/60 cmm.; bacteria, 285,300,000 per cc. The contrast between these counts and those in Table 11 shows clearly the effect of sanitary methods on the character of the product.

Investigation showed the following to be the principal causes for high counts of microorganisms in tomato pastes: (*a*) Partly decayed stock imperfectly washed or sorted; (*b*) delay at some stage in the

manufacturing process, allowing secondary spoilage to occur; and (*c*) the use of "swells" or spoiled "half stock" ("ministrella"). The particular condition causing the spoilage can sometimes be established in the finished product by microscopical examination. Sharp distinctions, however, can not at this stage of the investigations be drawn in every case.

TABLE 12.—*Analysis of Italian tomato sauces made from bad or questionable stock as determined by visual inspection.*[1]

Sample.	Description.	Character from inspection.[2]	Yeasts and spores.	Bacteria.	Fields with molds.
			Per 1/60 cmm.	*Million per cc.*	*Per cent.*
500	From bin-stored tomatoes in bad condition	C	1,120	466	96
501	From partly decayed stock	C	843	233	65
502	From tomatoes not washed or sorted; of doubtful character	(3)	114	92	24
503	From tomatoes not washed or sorted; not approved	C	315	220	11
516	From tomatoes not approved	C	160	72	14
517	From freshly delivered, but not washed, tomatoes	(3)	54	30	3
526	From tomatoes stored in bin overnight; partly decayed	C	440	200	38
527	From tomatoes held overnight in bin; objectionable conditions	C	230	96	39
534	From tomatoes stored in bin overnight; partly decayed	C	340	96	43
540b	From tomatoes stored in bins from Saturday till Monday; bad condition	C	900	360	100
545b	From tomatoes stored in bins 3 or 4 days; 50 per cent of fresh tomatoes added	C	1,200	700	60
547	From tomatoes stored in bin 24 hours; bad condition	C	750	290	60
550	From tomatoes not approved because of handling system	C	300	72	27
551b	From basket-stored tomatoes; rather sour and moldy	C	700	115	56
559	From bin-stored tomatoes; sour and moldy	C	218	108	31
562a	From trimmings allowed to stand; bad condition	C	580	1,250	72
574	From trimmings; bad condition	C	600	300	72
576	From trimmings and bad tomatoes	C	650	300	88
586	From partly decayed tomatoes	C	400	400	88
597	From tomatoes, many decayed	C	120	125	80
598	do	C	240	108	70
600	do	C	220	96	55
601	From objectionable trimmings	C	550	400	30
604	From partly decayed stock	C	375	200	54

[1] Analyses by B. J. Howard.
[2] C, condemned; bad product from inspection of raw material.
[3] Doubtful, or open to criticism.

The influence of improper storage and handling of stock is illustrated by two tests run the same day at one of the plants visited. Sample A was taken from tomatoes stored in a bin overnight and not sorted or washed before being made into sauce. Sample B was a sauce made at the same plant the same day as A but came from good stock promptly handled.

Sample.	Mold count.	Yeast and spore count.	Bacterial count.
A	43	340	96
B	12	48	24

Thus it is seen that high counts on this class of products, as well as on those less concentrated, indicate bad stock or insanitary handling.

METHOD FOR MICROANALYSIS OF TOMATO PRODUCTS.

Since the publication of Bureau of Chemistry Circular 68 no statement of the microscopical method used by the department has been issued. The bureau has received repeated requests for a restatement of the method, including more definite details of manipulation than were given in the circular. In 1915, after the method incorporating the most important of these details had been rewritten, the Association of Official Agricultural Chemists adopted it as a tentative method and published it November, 1916, in the Journal of the Association of Official Agricultural Chemists. At the meeting of the same association in November, 1916, a few minor changes were authorized. As it is uncertain when the association will publish the method in its amended form, permission has been granted by the chairman of the Board of Editors of the Association of Official Agricultural Chemists to incorporate it here.

49[1] APPARATUS.

(*a*) *Compound microscope.*—Equipped with apochromatic objectives and compensating oculars, giving magnifications of approximately 90, 180, and 500 diameters. These magnifications can be obtained by the use of 16 and 8 mm Zeiss apochromatic objectives with X6 and X18 Zeiss compensating oculars, or their equivalents, such as the Spencer 16 and 8 mm apochromatic objectives with Spencer X10 and X20 compensating oculars, the drawtube of the microscope being adjusted as directed below.

(*b*) *Thoma-Zeiss blood counting cell.*[2]

(*c*) *Howard mold counting cell.*—Constructed like a blood-counting cell but with the inner disk (which need not be ruled) about 19 mm in diameter.[2]

50[1] MOLDS (TENTATIVE).

Clean the special Howard cell so that Newton's rings are produced between the slide and the cover glass. Remove the cover and place, by means of a knife blade or scalpel, a small drop of the sample upon the central disk; spread the drop evenly over the disk and cover with the cover glass so as to give an even spread to the material. It is of the utmost importance that the drop be mixed thoroughly and spread evenly; otherwise the insoluble matter and consequently the molds are most abundant at the center of the drop. Squeezing out of the more liquid portions around the margin must be avoided. In a satisfactory mount Newton's rings should be apparent when finally mounted and none of the liquid should be drawn across the moat and under the cover glass.

Place the slide under the microscope and examine with a magnification of about 90 diameters and with such adjustment that each field of view represents approximately 1.5 sq. mm of area on the mount.[3] This area is of vital importance and may be obtained by adjusting the drawtube to the proper length as determined by actual measurement of the field, a 16 mm Zeiss apochromatic objective with a Zeiss X6 compensating ocular or a Spencer 16 mm apochromatic objective with a Spencer X10 compensating ocular, or their equivalents, being used to obtain the proper magnification.

---

[1] These numbers refer to the sections as given in the Journal of the Association of Official Chemists, November, 1916.

[2] Comment by authors: In using these cells the plane parallel cover glasses furnished with them by maker should be used instead of the ordinary microscope cover glasses, since the latter are subject to curvatures that introduce errors in the thickness of the mounts.

[3] Comment by authors: In order to have an area of 1.5 sq. mm the diameter of the microscopic field should be 1.382 mm. This is determined by using a stage micrometer and adjusting the length of the microscope drawtube. Obviously after the proper drawtube length has been secured that adjustment should be noted and always used in making mold counts.

Observe each field as to the presence or absence of mold filaments and note the result as positive or negative. Examine at least 50 fields, prepared from 2 or more mounts. No field should be considered positive unless the aggregate length of the filaments present exceeds approximately one-sixth of the diameter of the field. Calculate the proportion of positive fields from the results of the examination of all the observed fields and report as percentage of fields containing mold filaments.

51[1] YEASTS AND SPORES (TENTATIVE).

Fill a graduated cylinder with water to the 20 cc mark, and then add the sample till the level of the mixture reaches the 30 cc mark. Close the graduate, or pour the contents into an Erlenmeyer flask, and shake the mixture vigorously 15 to 20 seconds. To facilitate thorough mixing the mixture should not fill more than three-fourths of the container in which the shaking is performed. For tomato sauce or pastes or products running very high in the number of organisms, or of heavy consistency, 80 cc of water should be used with 10 cc or 10 grams of the sample. In the case of exceptionally thick or dry pastes it may be necessary to make an even greater dilution.

Pour the mixture into a beaker. Thoroughly clean the Thoma-Zeiss counting cell so as to give good Newton's rings. Stir thoroughly the contents of the beaker with a scalpel or knife blade, and then, after allowing to stand 3 to 5 seconds, remove a small drop and place upon the central disk of the Thoma-Zeiss counting cell and cover immediately with the cover glass, observing the same precautions in mounting the sample as given under 50. Allow the slide to stand not less than 10 minutes before beginning to make the count. Make the count with a magnification of about 180 to obtain which the following combinations, or their equivalents, should be employed: 8 mm Zeiss apochromatic objective with X6 Zeiss compensating ocular, or an 8 mm Spencer apochromatic objective with X10 Spencer compensating ocular with draw tube not extended.

Count the number of yeasts and spores [2] on one-half of the ruled squares on the disk (this amounts to counting the number in 8 of the blocks, each of which contains 25 of the small ruled squares). The total number thus obtained equals the number of organisms in 1/60 cmm if a dilution of 1 part of the sample with 2 parts of water is used. If a dilution of 1 part of the sample with 8 parts of water is used the number must be multiplied by 3. In making the counts the analyst should avoid counting an organism twice when it rests on a boundary line between 2 adjacent squares.

52[1] BACTERIA (TENTATIVE).

Estimate the bacteria from the mounted sample used in 51, but allow the sample to stand not less than 15 minutes after mounting before counting. Employ a magnification of about 500, which may be obtained by the use of an 8 mm Zeiss apochromatic objective with an X18 Zeiss compensating ocular with draw tube not extended, or an 8 mm Spencer apochromatic objective with an X20 Spencer compensating ocular with a tube length of 190, or their equivalents. Count and record the number of bacteria in a small area consisting of 5 of the small-sized squares. Move the slide to another portion of the field and count the number on another similar area. Count 5 such areas, preferably 1 from near each corner of the ruled portion of the slide and 1 from near the center. Determine the average number of bacteria per area and multiply by 2,400,000, which gives the number of bacteria per cc. If a dilution of 1 part of the sample with 8 parts of water instead of 1 part of the sample with 2 parts of water is used in making up the sample, then the total count obtained as above must be multiplied by 7,200,000. Omit the micrococci type of bacteria in making the count.

---

[1] These numbers refer to the sections as given in the Journal of the Association of Official Chemists, November, 1916.

[2] Comment by authors: The organisms counted as "yeasts and spores" are the yeast cells, and yeast and mold spores, not bacteria spores.

EXPLANATION OF CALCULATIONS.

Figure 5 has been prepared to make somewhat clearer the explanation of the areas denoting the yeast and spore and bacterial counts. The light lines in the figure show the arrangement of rulings on the entire slide. The squares (A, A, etc.) and rectangles (B, B, etc.) designated in the figure by the heavy lines indicate the portions used for the yeast and spore and for the bacterial counts, respectively. The 8 large squares, A, A, etc., are the squares used for yeast and spore counts. Each of these squares has 25 of the small squares. The sum of the organisms counted in the 8 squares marked A, A, etc., is the number in 1/60 cmm. if a dilution of one part of product to two parts of water is used.

Yeast and spore count.—The ruled square on the slide is 1 mm on each side and the cell is 1/10 mm deep. The volume of the ruled part is therefore 1/10 cmm. The ruled area is divided into 16 large squares and the number of organisms is counted in 8 of these, which is equivalent to 1/2 of 1/10 cmm, or 1/20 cmm. If a dilution of one part of the product to two parts of water is used 1/3 of 1/20 cmm, or 1/30 cmm as representing the actual amount of original stock in which organisms are counted, is obtained.

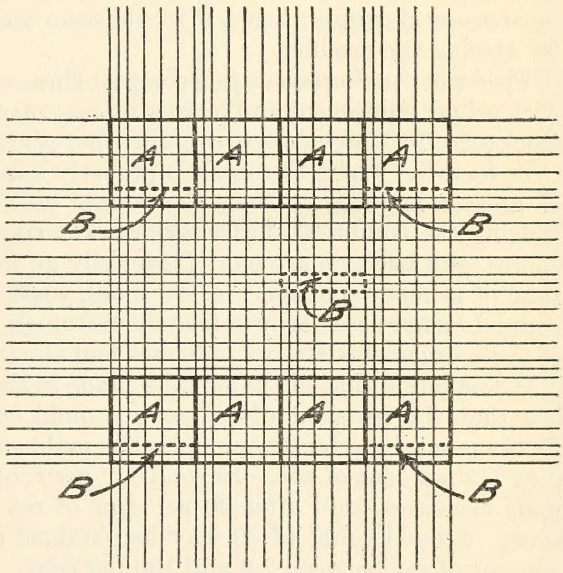

FIG. 5. Diagram of Thoma rulings. One millimeter divided by lines into 20 spaces in each direction, each space equaling 1/20 mm. To facilitate counting, every fifth space is subdivided by a line through the middle.

Bacterial count.—The rectangles, B, B, etc., each including 5 of the smallest squares, represent the areas, used in making the bacterial count. Similar rectangles of equal area might be selected, the object being to count 5 such areas well distributed over the ruled portion of the slide. The *average* number of bacteria counted on 5 rectangles, such as B, B, etc., multiplied by 2.4 million, equals the number of bacteria per cubic centimeter. In calculating the bacteria, it is observed that there are 400 (20×20) small squares on the slide. The number of bacteria in rectangles (B, B, etc.), each containing 5 of these small squares, are counted and an average made. This average represents the bacteria in 1/80 of the total ruled area. Since the cell is 1/10 mm deep, the volume represented by the organ-

isms counted is 1/80×1/10, or 1/800 cmm. With the usual dilution of one part of product to two parts of water the actual volume in which the number of organisms is determined is 1/3 of 1/800 cmm or 1/2400 cmm, or $\frac{1}{2,400,000}$ cc.

SUMMARY.

Tomato products promptly made from stock judged acceptable by visual inspection never show high counts of microorganisms. Similarly, products made from stock obviously not good or from stock improperly handled usually show high counts. It may therefore be assumed that high counts of organisms in such products indicate unmistakably that the stock used was in bad condition or was handled in an insanitary manner during manufacture.

It was found that tomato pulp stored in barrels usually gave high microscopical counts; hence it would seem inadvisable to use barrels for storing the product.

Field work performed during the past three seasons has proved that with proper equipment and factory management there is no reason for stock ready for the cyclone to contain over 1 per cent of decayed material.

In factories where the stock is properly handled the mold count is of greater importance than the counts on the other organisms in judging the condition of the raw stock. High counts of yeasts and spores, and bacteria are more frequently an indication of secondary than of primary spoilage. A low mold, yeast and spore, or bacterial count does not necessarily indicate sound stock, but a high count in any of these organisms always indicates bad stock or improper handling.

It was found that of the samples made in the laboratory none with less than 5.5 per cent of rot gave a mold count of more than 50. In the case of the factory samples the mold count rises sharply from 0 to 1/2 per cent of rot. Beyond 1/2 per cent the rate of rise gradually decreases, until after 20 per cent of rot the rate of increase is slow. A mold count of 40 may be obtained in samples having any amount of rot between 2.2 and 100 per cent.

A yeast and spore count of 20 represents about 1 per cent of decay. From this point the rate of increase is slow.

A bacterial count below 15,000,000 indicates little as to the amount of decay. Beyond this point, however, up to 20 per cent of rot the rate of increase is about 20,000,000 for each per cent of rot.

An investigation of the manufacture of tomato sauces and pastes in Italy showed that Italian products should be equal to American products made under similar conditions. The mold count for the concentrated products was found to be about the same as that for pulp, and the yeast and spore and bacterial counts to be proportional to the degree of concentration. Sauces and pastes made from objectionable material run particularly high in yeasts, spores, and bacteria. High counts on this class of products, then, indicate bad stock or insanitary handling.

○